## FECUNDITY OF SAN MARCOS SALAMANDERS IN CAPTIVITY

PAIGE A. NAJVAR,\* JOE N. FRIES, AND JOHN T. BACCUS

U.S. Fish and Wildlife Service, Austin Ecological Services, 10711 Burnet Road, Suite 200, Austin, TX 78758 (PAN)
U.S. Fish and Wildlife Service, San Marcos National Fish Hatchery and Technology Center, 500 East McCarty Lane,
San Marcos, TX 78666 (JNF)

Department of Biology, Texas State University at San Marcos, 601 University Drive, San Marcos, TX 78666 (JTB)

\*Correspondent: Paige\_Najvar@fws.gov

ABSTRACT—We placed 24 pairs of adult San Marcos salamanders in small flow-through aquaria with rocks and aquatic moss. Some aquaria had clear marbles within tubes through which water upwelled into each aquarium. Seven clutches (eggs per clutch: mean = 34.7; range = 2 to 73) were produced during a 9-month period. Overall hatching success was 29.6%. One breeding pair reproduced 3 times (eggs per clutch: mean = 44; range = 27 to 59) at an average of 64-day intervals during a 191-day period. Once mature, a single female potentially could produce 176 eggs per year in captivity.

RESUMEN—Veinticuatro pares de adultos de salamandras de San Marcos fueron colocados en pequeños acuarios de agua corriente con rocas y musgo acuático. Algunos acuarios tuvieron canicas claras dentro de tubos a través de los cuales el agua fluyó en cada acuario. Siete puestas (huevos por puesta: promedio = 34.7; rango = 2 a 73) fueron producidas durante un período de 9 meses. El éxito total de incubación fue de 29.6%. Un par de crianza se reprodujo 3 veces (huevos por puesta: promedio = 44; rango = 27 a 59) a un promedio de 64 días de intervalos durante un período de 191 días. Una vez madura, una hembra potencialmente puede producir 176 huevos por año en cautiverio.

The San Marcos salamander (Eurycea nana), a federally listed threatened species (U. S. Department of the Interior, 1980), is endemic to the headwaters of the San Marcos River, Hays County, Texas (Bishop, 1941; Chippindale et al., 1998). This species inhabits the constantly flowing waters of the San Marcos Springs issuing from the Edwards Aquifer through limestone faults along the Balcones Fault Zone. These springs fill Spring Lake at the headwaters of the San Marcos River (Brune, 1981). The range of E. nana includes Spring Lake and extends 150 m downstream of Spring Lake Dam (Nelson, 1993).

Previous studies addressing the natural history of the San Marcos salamander provided limited information about reproduction in this species. Both sexes become reproductively mature upon reaching 41 mm in total length (Bishop, 1941). Tupa and Davis (1976) asserted that male *E. nana* reach sexual maturity or possess at least one fully pigmented lobe in each testis after attaining 35 mm in total length. They also suggested that at any time females carry no more than about 20 eggs, 1.5 to 2.0 mm in diameter, in the abdominal region. However, Nelson (1993) found the number of eggs within females ranged from 23

to 37. Although juveniles and gravid females occur in Spring Lake during every month of the year (Bogart, 1967; Tupa and Davis, 1976), eggs of *E. nana* have never been observed in the wild. Observations of gravid females and juveniles suggest *E. nana* likely breeds throughout the year in the lake. We report additional natural history, primarily fecundity, observed while working with the San Marcos salamander in captivity.

We collected salamanders (total length about 20 to 50 mm, likely less than one year of age) from the San Marcos River in 1998 and 1999 and brought them to the National Fish Hatchery and Technology Center (NFHTC) in San Marcos, Texas. Individuals were segregated into 2 groups: salamanders visibly gravid and not visibly gravid. Each group was supplied with a mixture of recirculated water and fresh well-water maintained at approximately 22°C to mimic the temperature of their natural habitat. We examined each salamander for eggs monthly for 8 mo. Those carrying visible eggs were categorized as sexually mature females (Tupa and Davis, 1976) and were kept separate from those without eggs. Non-gravid salamanders greater than 41 mm in total length (Bishop, 1941) were

assumed to be sexually mature males and were the only males used. To maximize the likelihood that at least some eggs would be oviposited, we used salamanders with many eggs. Additionally, salamanders with physical anomalies (e.g., curvature of the spine, shrunken or swollen abdomen) were assumed to be unhealthy and were excluded.

Salamanders were maintained in 6-L (working volume) flow-through aquaria. A set of 8 aquaria was placed over each of three 442-L recirculation systems (a total of 24 aquaria) and provided with a naturally changing photoperiod. Four somewhat flat limestone rocks (about 5 to 10 cm in diameter) were placed in all aquaria and stacked in a pyramid arrangement. Aquatic moss (Amblystegium riparium) and native snails (both quarantined earlier for several months) were added to each aquarium in similar amounts. Upwelling flow tubes were attached to the bottom of half (12) of the aquaria. The tubes measured 30 cm in length and 5 cm in diameter, and contained clear marbles measuring 2.5 cm in diameter. Water upwelled through the tubes to simulate flow of water from spring openings common in the natural habitat of the salamander. Rocks, marbles, and moss provided cover and oviposition substrate, while the snails kept the aquarium glass free of algae and provided a constant food source for the salamanders. Additionally, pairs of salamanders were fed approximately equal amounts of commercially raised annelids 3 times per week and zooplankton, when available, to provide a diverse diet, because nutrition requirements are unknown for this species. Details of the set-up and culture methods are given in Najvar (2001). On 12 January 2000, one pair of San Marcos salamanders was placed in each of the 24 aquaria.

Salamander eggs produced by breeding pairs were removed from aquaria 3 d after first observation. A plastic baster was used to gently dislodge and suck the eggs from the substrate. They were placed on a plastic mesh substrate in a separate aquarium that received fresh wellwater and recirculated flow. The work was completed on 12 October 2000 and was covered under Federal permit number TE-676811-1 and State permit number SPR-0390-045.

Seven clutches (6 viable) produced a total of 243 eggs. Mean clutch size was 34.7 (range 2 to 73; SD = 23.5). All eggs were deposited between 3 March and 21 July 2000. The time between

oviposition and hatching ranged from 16 to 24 d, with an average of 21.3 d (SD = 2.6). Six clutches produced 72 larvae total. Overall hatching success was 29.6% (range 0 to 52.5%; SD = 19.8). The average number of hatchlings per clutch was 10.3. In aquaria equipped with upwelling-flow tubes, about half of the eggs were found in the aquatic moss, and half were found on marbles within the tubes, indicating that natural substrate is not necessary for oviposition in captivity.

One breeding pair reproduced 3 times during a 191-day period. These 3 clutches sequentially yielded 46, 59, and 27 eggs. This pair produced a total of 55 hatchlings. Fifty-one days elapsed between the introduction of the pair into the aquarium and the deposition of the first clutch, a 73-day period between the deposition of the first and second clutches, and a 67-day period between the second and third clutches. Newly developed eggs were observed in the abdomen of the female 24, 22, and 28 d after the first, second, and third reproductive events, respectively. The salamander was gravid with large eggs (estimated to be approximately 2 mm in diameter) 45 d following the deposition of the first clutch and 44 d following the deposition of the second clutch. Fifty-nine days after the third reproductive event, the female was gravid with eggs of many different sizes; however, these eggs were not deposited.

The maximum number of eggs (approximately 2 mm in diameter) per clutch (73) was higher than Nelson (1993) reported for E. nana. Four of 7 clutches exceeded the 20 eggs suggested by Tupa and Davis (1976). Three of 7 clutches exceeded 37 eggs found by Nelson (1993). Because no eggs of E. nana have been found in its habitat, it cannot be determined if clutch sizes of 73 occur in the wild. San Marcos salamanders have lived about 4 y at the NFHTC, and numerous eggs have been observed in captive females one year of age. Given these observations (132 eggs in 9 mo, 176 eggs per year), and presuming fecundity does not degrade appreciably over time, a single female salamander potentially could produce 500 or more eggs in her lifetime in captivity. Salamanders at the NFHTC were fed at least every other day, provided with adequate cover, and isolated from predators. This environment might have allowed female salamanders to devote more energy toward egg production in captivity than possible in the wild.

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